Possible Application of Urinary Analysis to Estimate Dissolution of Some Man-made Vitreous Fibers

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A preliminary study at the Institut National de l'Environnement Industriel et des Risques (INERIS) examined the dissolution of three man-made vitreous fiber samples (glasswool, rockwool, glass microfibers: JM 100) after intraperitoneal injections in male Wistar rats. The chemical composition of the original fibers was determined by inductively coupled plasma spectrometry (ICP). The urine of the rats was collected at fixed times between day 1 and day 204, and the ICP was used to look for elements known to be present in the original fibers. At day 204, a piece of omentum was removed at autopsy, ashed and analyzed by energy dispersive X-ray analysis (EDXA) to identify the elements remaining in the fibers. Silicon and aluminium were retained in the fibers from all samples at day 204. Losses in calcium, sodium, magnesium, and sulfur were observed, but these elements were not studied in the urine samples because they are naturally present in relatively high concentrations in rat cells and biological fluids. Although there was a loss of zinc from the glass microfibers, no corresponding difference was observed between the zinc levels excreted by the treated animals and by the controls. Similarly, despite the loss of manganese from the rockwool fibers at day 204, none was detectable in the urine samples. Titanium, present at the 0.3% level in rockwool, was not detectable by EDXA at day 204, but small quantities were detected in the first 2 weeks in the urine samples of rats treated with rockwool. The barium content of the retained glass microfibers (JM 100) had decreased at day 204, and this element was detectable in the corresponding urine samples. It was considered that Ti and Ba could be suitable biomarkers of exposure to rockwool and glass microfibers (JM 100). — Environ Health Perspect 102(Suppl 5):217–219 (1994)

Key words: man-made vitreous fibers, durability, exposure biological marker, urinary excretion, toxicology, intraperitoneal injections

Introduction

After inhalation, man-made vitreous fibers (MMVF) are deposited in the upper respiratory tract and the pulmonary alveoli. Certain fibers eventually are eliminated via bronchial or lymphatic action, though the time this takes may vary. Other fibers remain in the pulmonary fluids, undergoing transformation including fragmentation and partial dissolution (*1–12*). The chemical elements liberated by dissolution of fibers in the lungs can therefore spread throughout the organism before being eliminated (*13,14*), for example by urinary or biliary action.

If the elements normally are present in the organism in large quantities, as is the case for calcium, potassium, sodium, or magnesium, it is reasonable to assume that the additional contribution from inhaled fibers would be negligible; if the elements normally are present only in small quantities, any increase could prove harmful. At the same time, an increase in the urinary excretion of these elements could provide an indication of occupational exposure.

Most previous studies of natural or man-made vitreous fibers have focused on the transformation of the fibers or their by-products in the pulmonary system. Few authors have studied the other organs that might be affected (*15*).

In two epidemiological studies of MMVF workers, increased death rates due to bladder cancer (*16*) or to liver or bile duct cancer have been observed (*3*). The aim of this preliminary study was to explore the urinary excretion from rats that had received intraperitoneal injection of three MMVF samples.

Materials and Methods

The MMVF samples tested, were glasswool, rockwool and glass microfiber (JM 100). The chemical composition of the original fibers was determined quantitatively by inductively coupled plasma spectrometry (ICP) (Table 1). The fibers were ground prior to injection.

Table 1. Chemical composition of original fibers by ICP.

	Glasswool		Rockwool		JM 100		Oxides
Elements	% by weight	% as oxide	% by weight	% as oxide	% by weight	% as oxide	
Na	10.50	14.2	0.22	0.3	13.30	17.9	Na_2O
Mg	0.10	0.2	2.00	3.3	0.30	0.5	MgO
Al	1.60	3.0	5.80	11.0	2.80	5.3	Al_2O_3
Si	29.14	62.4	18.62	39.8	26.10	55.8	SiO_2
K	1.20	1.5	0.33	0.4	1.95	2.4	K_2O
Ca	5.90	8.3	27.80	38.9	1.90	2.7	CaO
Ti	0.03	0.1	0.30	0.5	—	—	TiO_2
Cr	—	—	0.01	—	—	—	Cr_2O_3
Mn	0.03	—	0.36	0.5	—	—	MnO
Fe	0.40	0.6	0.70	1.0	0.05	0.1	Fe_2O_3
Zn	—	—	—	—	2.60	3.2	ZnO
Sr	0.01	—	0.04	0.1	0.07	0.1	SrO_2
Ba	0.04	0.1	0.05	0.1	4.20	4.7	BaO
Total		90.4		95.6		92.7	

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Table 2. IPC determination in rat urine of MMVF elements at five times.

Fiber type	Days	Elements, mg/24-hr pooled urine Si	Ti	Mn	Zn	Ba
	1	351	[a]	[a]	7.3	[a]
	7	504	[a]	[a]	19.9	[a]
Glasswool	14	501	[a]	[a]	10.8	[a]
	28	389	[a]	[a]	9.2	[a]
	204	274	[a]	[a]	10.2	[a]
	1	490	3.4	[a]	26.6	[a]
	7	550	5.5	[a]	16.5	[a]
Rockwool	14	516	1.8	[a]	12.6	[a]
	28	583	[a]	[a]	15.1	[a]
	204	298	[a]	[a]	8.3	[a]
	1	535	[a]	[a]	8.1	9.2
	7	496	[a]	[a]	17.5	5.6
JM 100	14	455	[a]	[a]	20.0	5.0
	28	564	[a]	[a]	10.2	2.8
	204	357	[a]	[a]	11.9	1.4
Control		457 ± 106	[a]	[a]	10.3 ± 2.6	[a]

[a] Lower than detection limit.

Three groups of male Wistar rats (Strain ICO: WI–IOPS AF/H, Iffa Credo, L'Arbresle, France) received an intraperitoneal injection of 50 mg MMVF in 2 ml saline. A fourth control group of 4 rats received saline alone. The animals were housed in a protected zone. The 24 hr urine samples from the animals were collected at days 1, 7, 14, 28, 204 after injection. The urines were weighed and frozen. At day 204, the animals were sacrificed and a piece of omentum was preserved. A macroscopic examination of the urinary tract was performed. The urine samples were dried and ashed, and the principal elements of the original fibers were determined by ICP. The omentum sample was ashed at low temperature, and the fibers present were examined by transmission electron microscopy (TEM) equipped with an energy dispersive X-ray analysis unit (EDXA). Spectra were compared with those of the original fibers.

Results

The three fiber types recovered from the omentum samples after residing for 204 days in the peritoneal cavity were examined by EDXA.

Glasswool showed an increased K content and had retained Al and Si, but had lost Na and Ca. Rockwool showed an increased K content and had retained Al and Si, but had lost Mg, Ca, S, and Mn. It was not possible to measure Ti. Glass microfiber (JM 100) retained Al, Si, and K, but lost Ba, Na, C, and Zn. Si, Ti, Mn, Zn, and Ba were determined by ICP in the urine samples (Table 2). Macroscopic examination of the urinary tract showed a kidney tumor in one animal treated with glasswool, but this was not significant because of the small number of rats in each group.

Discussion

Al and Si were present in the omentum samples taken at day 204 from the rats treated with all three fiber types, and were looked for in the urine samples. Si was excreted in considerable amounts in urines of both controls and treated animals, which would mask any slight increase due to fiber dissolution. Si, therefore, could not be used as a biological marker of exposure. Ca was low in all fibers retained at day 204; but because the Ca concentration in biological fluids is very high, the contribution from dissolved fibers to the total Ca pool would be negligible. Moreover, Ca urinary excretion is unimportant in the metabolic regulation of Ca. Relatively few elements were found by EDXA of retained glasswool fibers. Although Na was lost from the residual fibers, it could not be used as a biomarker for the same reason that Ca could not be used.

At day 204, the residual rockwool fibers showed reduced amounts of Mg, S, and Mn, but neither Mg nor S could be used as biomarkers because of their natural metabolism. However, their presence could be significant since these two elements could become oxidized during dissolution of deposited fibers in the lung, and their oxides would be irritants of the mucous membranes (*17*). Mn was not detected in the urine samples. Nevertheless, it might still be a suitable marker of exposure, since its storage and excretion are primarily linked to hepatic activity (*17*).

ICP analysis of rockwool indicated that Ti was present only to the extent of 0.3%, and it was not quantifiable by EDXA in the residual fibers at day 204. At days 1, 7, and 14, small amounts of Ti were found in the urine samples from rockwool treated rats, but this result has to be checked because of the low level of Ti present in the fibers and the small number of animals used in the study. Ti, even when oxidized, is considered to be toxicologically inactive; therefore it should be considered a possible exposure marker.

The reduction of Na in the residual glass microfiber (JM 100) again cannot be used as a biomarker because of the negligible contribution it would make to the Na pool; Zn loss cannot be used because urinary excretion was the same both in control and treated groups.

The EDXA showed a reduction of Ba in the residual glass microfibers (JM 100) and this element was excreted in the urine of rats that had been injected with these microfibers. These results suggest that barium excretion might be a suitable biomarker of fiber exposure.

It should also be noted that soluble Ba salts are highly toxic and can cause extensive hypokalemia. EDXA of the samples of residual glasswool and rockwool both showed an increase in K at day 204; in contrast, the EDXA of the JM 100 samples, which contained Ba, showed no change in K content, suggesting a parallel with the hypokalemia phenomenon.

Conclusion

The "dissolution" of fibers could give rise to a number of complex biological mechanisms in which the different elements present in the fibers could be involved. The way that the presence of Ba in a fiber residing in the peritoneal cavity affects the level of K is a good example of this interaction.

It would be worthwhile to extend these preliminary studies, using intratracheal administration or inhalation of fibers and to explore the metabolism of the different component elements to determine the possible formation of toxic compounds and their routes of elimination via liver, kidney, or digestive tract.

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